Welcome to biological activity of antibodies. We probably did more than you ever wanted to know about antibody structure, and now it is time to go actually look at them in action. I think of an antibody as being like a cross between a kick me sign and a smart bomb. So, just to remind you what we're starting with, here is my antibody model. We have, again, the arms with the recognition regions in the end, and here and here, and we have the FC stem, and we're going to look at how some of the specifics of the stem will work, when the antibody is in action. But first, we want to look at the specific way that this antibody binds to antigen. If you look at it closely, you can see we have the six CDR loops, three on each of the arms of the antibody. In this sense, they're like fingers. Well, I don't have six fingers but I do have five. So, let me put this antibody up here as a reminder and take up a virus. This is a virus that has a protein capsid on the exterior, and if an antibody recognizes it, those three loops will find some confirmation on the antigen, that is the protein of the virus, that those loops do something to sort of fit. Now, my favorite way of looking at the interaction between the stem are, excuse me, between the arms of the antibody and the virus is as a sort of form of enzyme substrate interaction, so that if I've got the loops of the antibody here, they are going to fit into some protein like this. So, here would be a region of the protein. This protein would be the epitope, that is this part of the protein would be the region that the antibody recognizes. We would have something in here that can involve very much like an enzyme substrate interaction, and then we can have forms of induced fit, and we will have charge, a minus charge here and a plus charge here, we'll it tell to put them together. But, in general, what you've got is a series of weak interactions that sum up to a very sort of, if it works well, tight fit between the arm and the antigen. So, this allows this antibody to recognize very clearly a very particular structure and that's the good news. Okay? The bad news is that you don't want that structure to be one of your own structures. So, one thing that determines whether you make an antibody to something is whether or not this is foreign. So, we want something here that is definitely not you and we have a series of tolerance measures that we put into place to make sure that I don't make an antibody against one of your own proteins. Sometimes they fail. Another thing that has to happen is that, for this to work, we'll see a receptor on one part of the cell, and then a receptor for a similar one on a different part of the cell and they must cross-link. So, there's a certain optimal size for these things. If they're not big enough, they won't be immunogenic. Another thing that happens is, that we need to have, we'll see later on, a second signal that goes into the cell when the B cell initially recognizes an antigen in order to set off an immunogenic reaction. So, that second cell, it can be from a tow-like receptor or danger signal. So, that is why we have adjevents when we have various kinds of vaccines. So, if a vaccine just presents a protein to a B cell without some sort of additional signal that this protein represents to danger, that vaccine is not going to work and it might even make a person tolerant to that protein. Now, the third issue we want to look at here is, what if we want to make an antibody to something that just doesn't cooperate? That is, it's too small. It's just not going to cross-link two of the receptors on the surface of a B cell. We call such potential epitopes haptens and a hapten is something that is too small to cross-link two receptors in a B cell. But, sometimes you want antibodies do that. At the end of this lecture, we'll look at monoclonal antibodies. That is we often want to make a whole pot load of antibodies that specifically recognize some kind of molecule and sometimes that molecule is pretty small. So, what are we going to do to fool the T cell into cooperating and giving us antibodies to a small but interesting molecule? Why would you want to do that? Well, some environmental pollutants are very small. Some poisons are very small. There are also drugs in people's system. So, if you want to do a drug test, a really specific drug test, that drug might be very small. So, the good news is, you can make antibodies to a small drug molecule by doing the following kind of manipulation. Haptens are not inherently immunogenic. You must work to get your body to sensitize to them and produce antibodies against them. So, to do this, we're going to show you that one of the basic techniques that used to get a small molecule to trigger an immune response. Now, that small molecule could be an environmental pollutant, a small hormone, or metal ion and it's something that ordinarily you wouldn't produce antibodies against. Say, it's something like Analin. I could inject you with enough Analin to kill you

and you would still never make an antibody against it. So, what I'm going to do is shrink this thing down, make a bunch of them, and then, I'm going to stick these small molecules on enlarged proteins, such as bovine, serum, albumin or BSA, and that's a protein that's extracted from tablet. So, here are the haptens and here they are attached to the BSA. Looks a little like a chocolate chip cookie, right? When I do this, when I basically doing, is nailing down the haptens, so that, they're not just floating around. So, I can say inject this complex into a rabbit and the rabbit will make antibodies against it. One thing the rabbit will make antibodies against is the BSA, that's a foreign protein. So, if I have a B cell where the recognition regions on the arms recognize an epitope on the BSA, I'm going to make antibodies to BSA. Now, on the other hand, I can also make antibodies to the junction where the hapten and the BSA come together. If you will recall, one thing that you can get is a beast sensitization to, say a region where two viral proteins come together, that can be an epitope, the junction of these two proteins and I can get that here. But, that's not what I want. What I really want is an antibody that recognizes the hapten and only the hapten all by itself. Here, we have one. This B cell just happens to have a recognition region that can bind to the hapten all by itself, because the hapten is stuck in the BSA. It will crosslink the two receptors in the B cell, that will set off a chain of developmental events and eventually that B cell will turn into a plasma cell, and make antibody to the hapten. Now, we're going to look back at this issue of how that whole activation process works a little later on in this lecture. But, for now, I'm just going to wave my hand and say this B cell is making lots and lots of antibody to that hapten. The cell couldn't have done that if those haptens hadn't been stuck on the BSA, because even though the antibody might have recognized the hapten, the antibody still wouldn't have been cross-link and we'll see that cross-linking is necessary for the activation process. Now, this B cell is eventually going to divide and produce lots of antibodies. So, here we have a situation where we have lots of little haptens might be in the air as a pollutant, or water as some kind of poison, or we might be looking for a drug in the human serum. In any event, we have now a way of picking these up. That is, we can make an antibody that recognizes this hapten all by itself. Here, it's binding to two of them. Here it's just one and now, two. So, in this case, if I mix these antibodies with the water or serum, or whatever I'm trying to test, then I can precipitate the antibodies out, I can see whether or not that hapten is present. So, I can use these antibodies that I developed in an artificial system to look for hapten out in the real world or in somebody's body and that gives us a real tool to examine and test our outside world.